Cortisol Secretion, Sensitivity, and Activity Are Associated With Hypertension in Postmenopausal Eucortisolemic Women

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Context: Previous data suggest a possible association between type 2 diabetes (T2D) and fragility fractures (FX) with the degree of glucocorticoid suppressibility (GCS) and peripheral activation or sensitivity even in persons without hypercortisolemia.

Objective: To investigate whether the degree of GCS, GC sensitivity, and peripheral activation in persons without overt or mild hypercortisolism are associated with hypertension and with the number of the possible consequences of cortisol excess among patients with T2D, fragility FX, and hypertension.

Design: Case-control study.

Setting: Outpatient clinic.

Patients: A total of 216 postmenopausal women without hypercortisolemia (age, 50 to 80 years; 108 with hypertension); 68 and 99 patients had fragility FX and T2D, respectively

Main outcome measures: We assessed 24-hour urinary free cortisol (UFF), cortisone (UFE), their ratio (R-UFF/UFE), (F-1mgDST), and the GC receptor *N363S* single-nucleotide polymorphism (*N363S*-SNP).

Results: Hypertension was associated with F-1 mgDST [odds ratio (OR), 3.3; 95% CI, 1.5 to 7.5; $P = 0.004$) and R-UFF/UFE (OR, 101.7; 95% CI, 2.6 to 4004.1; $P = 0.014$), regardless of age, body mass index, and presence of the N363S single nucleotide polymorphism and of T2D. The progressive increase in the number of possible consequences of cortisol excess was significantly associated with F-1mgDST levels ($R^2 = 0.125$; $P = 0.04$), R-UFF/UFE ($R^2 = 0.46$; $P = 0.02$), and the prevalence of N363S heterozygous variant (T = 0.46; $P = 0.015$), after adjustment for age.

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Abbreviations: 11βHSD, 11β-hydroxysteroid dehydrogenase; BMD, bone mineral density; BMI, body mass index; F-1mgDST, cortisol after a 1-mg overnight dexamethasone test; FX, fractures; GC, glucocorticoid; GCS, glucocorticoid suppressibility; GR, glucorticoid receptor; OR, odds ratio; R-UFF/UFE, ratio of urinary free cortison to urinary free cortisol; T2D, type 2 diabetes; UFE, urinary free cortisone; UFF, urinary free cortisol.

Conclusions: In postmenopausal women without hypercortisolemia, hypertension is associated with GCS and GC peripheral activation. The number of possible consequences of cortisol excess (among patients with hypertension, T2D, and fragility FX) is associated with GCS, GC peripheral activation, and the prevalence of the N363S heterozygous variant. (***J Clin Endocrinol Metab*** **104: 4441–4448, 2019)**

It is well known that glucocorticoid (GC) excess leads to specific signs (such as easy bruising, moon facies, buffalo hump, muscle atrophy, and striae rubrae), which are the clinical hallmarks of Cushing syndrome (1). It is also known that both clinically overt and asymptomatic hypercortisolism cases (frequently found in patients with incidentally discovered adrenal masses) are also associated with a higher prevalence of conditions not specific to cortisol excess, such as hypertension, type 2 diabetes (T2D), and fragility fractures (FX) (2–4).

However, some recent studies suggested that even in patients with normal cortisol levels, the degree of cortisol secretion can influence skeletal health and glucose metabolism. Indeed, cortisol levels appear associated with bone mineral density (BMD) in women with postmenopausal osteoporosis (5, 6). In addition, in eucortisolemic patients, the activity of the 11β-hydroxysteroid dehydrogenase (11βHSD) shuttle, which regulates GC peripheral activation by interconverting the inactive cortisol into active cortisone and vice versa (7), seems to influence the severity of osteoporosis (8) and the risk of vertebral fractures (9). Finally, the different GC receptor (GR) polymorphisms, which are thought to increase GC sensitivity, such as N363S (10), have been suggested to be associated with the FX risk in patients with no evidence of cortisol excess (11). In keeping with these findings, cortisol levels, although within the normal range, have been increased in patients with T2D compared with controls without T2D (12, 13). Moreover, both the BclI and N363S polymorphisms of the *GR* gene increase GC sensitivity *in vivo* and are associated with a worse glycometabolic and lipid profile (10, 14, 15). Finally, the activity of the 11βHSD enzymes is considered to play a role in glucose homeostasis in healthy persons (16), and in patients with T2D the modulation of 11βHSD activity may ameliorate diabetes control and hypertension (17, 18).

To date, the role of GC suppressibility (GCS), sensitivity, and peripheral activation in patients with hypertension is largely unknown. Moreover, given the well-recognized influence of GC on glucose metabolism and skeletal health, it is possible to conceive that, even in eucortisolemic patients, the degree of GCS and GC peripheral activation and sensitivity may be associated with the number of possible consequences among T2D, fragility FX, and hypertension. Therefore, the aim of the current study was to investigate, in patients with normal cortisol levels, whether the degree of GCS, GC sensitivity, and GC peripheral activation is associated with hypertension and the number of consequences among patients with fragility FX, T2D, and hypertension.

Patients and Methods

Patients

The current study is based on the population of a previous study designed to assess the possible association between cortisol secretion, 11βHSD activity, and the different polymorphisms of the *GR* gene and the presence of fragility FX in postmenopausal female patients with T2D (19). In that study, conducted from September 2011 through September 2014, 120 consecutive patients with T2D referred to our outpatient clinic for diabetes and 120 age-matched nondiabetic persons were enrolled. We chose to include only white persons to avoid biases related to the effect of the different races on vitamin D metabolism, bone, and vascular health (20). Patients with T2D were selected on the basis of the following criteria: age 50 to 85 years, postmenopausal status, age at T2D diagnosis older than 30 years; body mass index (BMI) of 19 to 40 kg/m^2, no insulin therapy during the first 2 years of the disease, and hemoglobin A1c $\leq$ 8.0%.

The exclusion criteria were signs of hypercortisolism (moon facies, striae rubrae, hypertrichosis, skin atrophy, and buffalo hump); history of ketoacidosis or hypoglycemia in the 6 months before enrollment; past or present therapy with glitazones, glucocorticoids (>3 months and/or >5 mg/d of prednisone equivalents), antidepressants, bisphosphonates, strontium ranelate, parathyroid hormone (1-34 and 1-84), denosumab, anticonvulsants, estrogens, or selective estrogen receptor modulators; presence of hyperthyroidism, rheumatoid arthritis, scleroderma, malabsorption, neoplasia, hyperandrogenism, alcoholism, depression, chronic renal failure, acute illnesses, or alterations of the sleep-wake cycle; or presence of proliferative or laser-treated retinopathy, overt diabetic nephropathy (macroalbuminuria> 300 mg/24 hours), or severe macroangiopathy (history of myocardial infarction; coronary artery bypass graft surgery; or percutaneous transluminal coronary, carotid, femoral, or femoral-popliteal angioplasty).

The persons without diabetes were consecutively recruited on the basis of the same inclusion and exclusion criteria from our outpatient clinic for endocrine diseases, where they were followed up for nonmorbid obesity, euthyroid nodular goiter, or euthyroid chronic lymphocytic thyroiditis.

No patients were treated with glucagon-like peptide-1 receptor agonists or other hypoglycemic agents that might have influenced blood pressure. The study was eventually performed among 99 patients with T2D and 107 without T2D (the latter of

which served as the control group); 21 diabetic and 13 nondiabetic patients did not complete the study protocol and were excluded from the analysis. In that previous study, 68 patients had fragility FX.

All participants provided informed consent, and the Milan (Area A) Ethic Committee approved the study.

In the present analysis, we pooled patients with T2D and nondiabetic controls, resulting in a single population of 206 postmenopausal women. In the whole population, we decided to compare data regarding cortisol secretion, 11βHSD activity, and prevalence of different polymorphisms of the *GR* gene in patients with and without hypertension. Therefore, we subdivided the whole sample into patients with hypertension (n = 108) and those without hypertension (n = 98).

Methods

In all patients, blood venous samples were taken in the morning after 10-hour fasting. All blood and urine samples were immediately stored at −20°C until analysis.

The cortisol secretion was evaluated by measuring morning plasma ACTH [normal values, 10 to 55 pg/mL (2.2 to 12.1 pmol/L)], 24-hour urinary free cortisol [UFF; normal values, 3 to 43 μg/24 hours (8.3 to 118.7 nmol/24 hours] and cortisone [UFE; normal values, 15 to 122 μg/24 hours (41.6 to 337.9 mol/24 hours], serum cortisol at 09:00 after the administration of 1 mg dexamethasone at 11 PM on the previous day [F-1mgDST; normal values < 1.8 μg/dL (50 nmol/L)].

At study entry, plasma morning ACTH levels (mean of three determinations at 20-minute intervals) and serum cortisol levels (after dichloromethane extraction) were measured by immunoradiometric assay (Brahms Diagnostica GmbH, Berlin, Germany) and by immunofluorometric assay (TDX-FLX, Abbott Diagnostika, Wiesbaden, Germany). The intra- and interassay coefficients of variation were ≤10% for ACTH and <3% for cortisol.

The UFF/UFE ratio (R-UFF/UFE) was used as a measure of 11βHSD type 2 activity. By inactivating cortisol to cortisone, this enzyme may modulate tissue exposure to active GC (7). UFF and UFE were determined by using liquid chromatography-tandem mass spectrometry after purification by an online TurboFlow system using a Cyclone column (ThermoFisher Scientific, Rodano, Italy). Cortisol and cortisone were separated during liquid chromatography (Hypersil Gold, ThermoFisher Scientific). Detection and quantification were performed by a triple quadrupole mass spectrometer (TSQ Quantum Access, ThermoFisher Scientific). The coefficient of variation is <10%, the accuracy is between 98% and 100%, and the limit of quantification is 1 μg/L for both cortisol and cortisone (21).

Genomic DNA was isolated from peripheral blood leukocytes using Illustra DNA Extraction Kit BACC2 (GE Healthcare, Little Chalfont, Buckinghamshire, United Kingdom). Detection of the BclI and N363S polymorphisms was assessed by polymerase chain reaction (PCR), as previously described (11).

Hypertension was defined as systolic blood pressure > 140 mm Hg diastolic blood pressure > 90 mm Hg, and/or receipt of antihypertensive treatment (22). T2D was diagnosed by using World Health Organization criteria (23). In all patients, the history of fragility FX (humerus, vertebrae, hip, and wrist) was recorded and verified by obtaining the medical reports. A conventional spinal radiograph in lateral (T4 to L4) and antero-posterior projection was obtained in all patients by using standardized techniques. Two trained radiologists, blinded to BMD and hormonal data, independently reviewed the radiographs. Vertebral FX were diagnosed on visual inspection by using semiquantitative visual assessment and defined as reductions of >20% in anterior, middle, or posterior vertebral height (24). A fracture was considered to be due to skeletal fragility if it was determined by mechanical forces that would not ordinarily result in fracture, known as low-level (or low energy) trauma (25)

Statistical analysis

In the previous study (17), the true difference in F-1mgDST levels between patients with and those without T2D was 0.2 (SD, 0.4). On the basis of those data, we would have needed to study 85 patients with hypertension and 85 without hypertension to be able to reject the null hypothesis that the population means of the patient and control groups were equal with a probability (power) of 0.9 (type I error α value of 0.05).

Statistical analysis was performed by using SPSS software, version 23.0 (IBM, Milan, Italy). The results are expressed as mean ± SD if not otherwise specified. The normality of distribution was tested by the Kolmogorov-Smirnov test. All variables were normally distributed.

Categorical variables were compared by using the χ^2 test or Fisher exact test, as appropriate. We compared continuous variables between patients with and those without hypertension by using the Student *t* test or Mann-Whitney *U* test for non–normally distributed variables. The comparisons among patients with no, one, two, or three possible consequences of cortisol excess among those with hypertension, T2D, and fragility FX were performed by using one-way ANOVA and Bonferroni *post hoc* analysis. The general linear model has been performed for adjusting the comparisons of ACTH, F-1mgDST, UFF, UFE, and R-UFF/UFE for the differences in age and BMI.

The logistic regression analysis assessed the association between the presence of hypertension (as dependent variables) with the cortisol-related parameters, after adjustment for age, BMI, and presence of T2D. The same analysis was used to assess whether the simultaneous presence of T2D, hypertension, and fragility FX was independently associated with the cortisol-related parameters after adjustment for age and BMI.

P values < 0.05 were considered to indicate statistically significant differences.

Results

Association between hypertension and degree of GCS, GC sensitivity, and GC peripheral activation

Data from the whole patient sample have been analyzed by comparing patients who have hypertension with those who do not have hypertension. The characteristics of these groups are reported in Table 1.

Age and BMI were greater in patients with hypertension than in those without hypertension. The prevalence rates of fragility FX, BclI and N363S polymorphic variants of the *GR* gene, and ACTH and UFF levels were similar between patients with or without hypertension

(Table 1). Patients with hypertension showed higher F-1mgDST levels, lower UFE levels, and higher R-UFF/UFE and frequency of T2D compared with patients without hypertension, even after adjustment for age and BMI by a general linear model. The age- and BMI-adjusted difference in R-UFF/UFE levels between patients with hypertension and those without hypertension was confirmed even after exclusion of patients with T2D from the analysis. At variance, after exclusion of patients with T2D, the difference in F-1mgDST levels did not reach statistical significance (Table 2).

However, in the whole group of patients, logistic regression analysis showed that hypertension was associated with F-1mgDST [odds ratio (OR), 3.29; 95% CI, 1.45 to 7.46; $P = 0.004$)], *R*-UFF/UFE (OR, 125.9; 95% CI, 3.29 to 4909.4; $P = 0.01$), age (OR, 1.06; 95% CI, 1.01 to 1.11; $P = 0.022$), and BMI (OR, 1.1; 95% CI, 1.02 to 1.18; $P = 0.01$) after adjustment for the presence of T2D (OR, 1.73; 95% CI, 0.89 to 3.36; $P = 0.109$) and the presence of the N363S polymorphic variant (OR, 1.12; 95% CI, 0.29 to 4.38; $P = 0.874$).

Association between GCS, GC sensitivity, and GC peripheral activation is associated with number of possible consequences of cortisol excess among patients with fragility FX, T2D, and hypertension

The progressive increase in the number of the possible consequences of cortisol excess (among those with hypertension, T2D, and fragility FX) was significantly associated with F-1mgDST levels ($R^2 = 0.125$; $P = 0.04$), with R-UFF/UFE ($R^2 = 0.46$; $P = 0.02$) and with the prevalence of the N363S heterozygous variant (T = 0.46; $P = 0.015$), after adjustment for age (Fig. 1).

The patients simultaneously affected with T2D, hypertension, and fragility FX were significantly older than patients without these conditions. Age-adjusted F-1mgDST and R-UFF/UFE and the prevalence of N363S heterozygous variant were higher in patients simultaneously affected with T2D, hypertension, and fragility FX than in those without (Table 3). The logistic regression analysis showed that the simultaneous presence of T2D, hypertension, and fragility FX was independently associated with F-1mgDST (OR, 5; 95% CI, 1.6 to 15.2; $P = 0.005$), R-UFF/UFE (OR, 131.6; 95% CI, 2.3 to 7417.8), and prevalence of N363S heterozygous variant (OR, 12.4; 95% CI, 2.9 to 53.1) after adjustment for age and BMI.

Discussion

The current study suggests that in postmenopausal women without hypercortisolism, hypertension is associated with GCS and GC peripheral activation (as mirrored by F-1mgDST levels and R-UFF/UFE, respectively) regardless of T2D, age, and BMI. In addition, we found that the progressive increase of the number of possible consequences of cortisol excess (among patients with T2D, hypertension, and fragility FX) (Fig. 1) was associated with cortisol secretion, GC peripheral activation, and GC sensitivity.

Table 1. Characteristics of Patients With and Without Hypertension

Characteristic	Patients With Hypertension (n = 108)	Patients Without Hypertension (n = 98)	*P* Value
Age, y	67.3 ± 7.4 (52–80)	62.4 ± 7.9 (37–80)	0.0001
BMI, kg/m^2	30.1 ± 4.9 (20.9–40.0)	27.8 ± 4.3 (19.9–39.1)	0.0001
Patients with diabetes, n (%)	64 (64.6)	35 (35.7)	0.001
Patients with fragility Fx, n (%)	31 (28.7)	23 (23.5)	0.39
ACTH, pg/mL[a]	16.8 ± 11.1 (5.0–55.0)	16.4 ± 9.1 (5.0–54.2)	0.711
F-1mgDST, μg/dL[a]	1.25 ± 0.43 (0.1–1.8)	1.02 ± 0.41 (0.4–1.8)	0.003
UFF, μg/24 h[a]	19.4 ± 8.9 (4.2–54.7)	20.2 ± 9.7 (5–53.1)	0.36
UFE, μg/24 h[a]	86.8 ± 20.6 (33.7–125)	94.7 ± 21.6 (37.1–166.3)	0.009
R-UFF/UFE[a]	0.24 ± 0.11 (0.06–0.6)	0.20 ± 0.08 (0.07–0.5)	0.008
N363S, n (%)			0.542
Wild type	99 (91.7)	92 (93.9)	
Heterozygous variant	9 (8.3)	6 (6.1)	
Homozygous variant	0 (0)	0 (0)	
Bcl1, n (%)			0.312
Wild type	66 (61.1)	50 (51)	
Heterozygous variant	38 (35.2)	42 (42.9)	
Homozygous variant	4 (3.7)	6 (6.1)	

Data are mean ± SD (range) or absolute number (%). Normal ACTH: 5 to 55 pg/mL; normal F-1mgDST: <1.8 μg/dL); normal UFE: 15 to 122 μg/24 hours); normal UFF: 3 to 43 μg/24 hours.

[a] After adjustment for age and BMI by general linear model.

Table 2. Characteristics of Patients With and Without Hypertension After Exclusion of Patients With Type 2 Diabetes

Parameters	Patients With Hypertension (n = 44)	Patient Without Hypertension (n = 63)	*P* Value
Age, y	67.3 ± 7.4 (52–80)	62.4 ± 7.9 (37–80)	0.0001
BMI, kg/m^2	30.1 ± 4.9 (20.9–40.0)	27.8 ± 4.3 (19.9–39.1)	0.0001
F-1mgDST, μg/dL[a]	1.18 ± 0.37 (0.1–1.8)	0.98 ± 0.37 (0.4–1.8)	0.22
R-UFF/UFE[a]	0.25 ± 0.96 (0.06–0.6)	0.19 ± 0.08 (0.07–0.5)	0.008
N363S, n (%)			0.99
Wild type	41 (93.2)	59 (93.7)	
Heterozygous variant	3 (6.8)	4 (6.3)	
Homozygous variant	0 (0)	0 (0)	
BclI, n (%)			0.631
Wild type	24 (54.5)	31 (61.7)	
Heterozygous variant	18 (40.9)	29 (46.0)	
Homozygous variant	2 (4.5)	3 (4.8)	

Data are mean ± SD (range) or absolute number (%). Normal F-1mgDST: <1.8 μg/dL.

[a] After adjustment for age and BMI by general linear model.

The possible influence of a different degree of cortisol secretion, peripheral activation, and sensitivity on bone and glucose metabolism has already been suggested (26, 27). Indeed, even in eucortisolemic patients, the genetic polymorphisms in 11βHSD enzymes correlate with the postdexamethasone cortisol levels, BMD (6), and fragility FX (9), and cortisol secretion may influence BMD (6) and the rate of bone loss in healthy adults (5). In keeping with these findings, cortisol levels, although within the normal range, have been increased in patients with T2D compared with controls without T2D (12, 13). Moreover, both the BclI and N363S polymorphisms of the *GR* gene increase GC sensitivity *in vivo* and are associated with worse glycometabolic and lipid profiles (10, 14, 15). Finally, the activity of the 11βHSD enzymes is considered to play a role in glucose homeostasis in healthy persons (16), and in patients with T2D the modulation of the 11βHSD activity has been suggested to ameliorate diabetes control and hypertension (17, 18).

At variance, the possible influence of the different degree of cortisol secretion, peripheral activation and sensitivity on hypertension has been scarcely investigated. Indeed, the idea that cortisol sensitivity and peripheral activation may play a role possible contributing factors in the pathogenesis of hypertension has been proposed by several authors on the basis of studies on both *GR* polymorphisms (28) and 11βHSD activity (29) and on the fact that hypertension prevalence is higher in patients with slightly increased cortisol secretion (30). However, to our knowledge, no studies to date have investigated, in a cohort of patients with normal cortisol levels, whether a different degree of GC secretion, peripheral activation, and sensitivity may be associated with the presence of hypertension. Our finding that postmenopausal patients with hypertension have increased cortisol secretion and peripheral activation might be important in the development of drugs designed to modulate cortisol secretion and/or peripheral activation for treating hypertension. All our patients were eucortisolemic, and, therefore, the real clinical relevance of the present data must still be determined. However, the fact that in patients with T2D inadequately controlled by metformin monotherapy 11βHSD type 1 inhibition improved hyperglycemia reinforces the possibility that even patients with hypertension may take advantage of drugs modulating 11βHSD activity (17).

This idea may be also suggested by the fact that, at least in our sample of postmenopausal women, the progressive increase in the number of possible consequences of cortisol excess among patients with T2D, hypertension, and fragility FX (Fig. 1) and that their simultaneous presence is associated with GCS, GC peripheral activation, and GC sensitivity regardless of age and BMI. This finding is in keeping with data from patients with adrenal adenomas in whom cortisol secretion was associated with the number of chronic complications (31, 32). If this finding, which is novel in persons without cortisol hypersecretion, were to be confirmed in a larger sample of patients, it would be possible to hypothesize that a single drug modulating cortisol secretion or GC peripheral sensitivity may simultaneously prevent hypertension, T2D, and osteoporosis. It is also of interest that in the current study, after adjustment for cortisol parameters, hypertension was unexpectedly weakly associated with age and BMI, as evidenced by both the low OR and the lower limit of the CI just above 1, reinforcing the idea of an independent role of cortisol in hypertension.

These data may have two main clinical implications. First, if in eucortisolemic patients the degree of

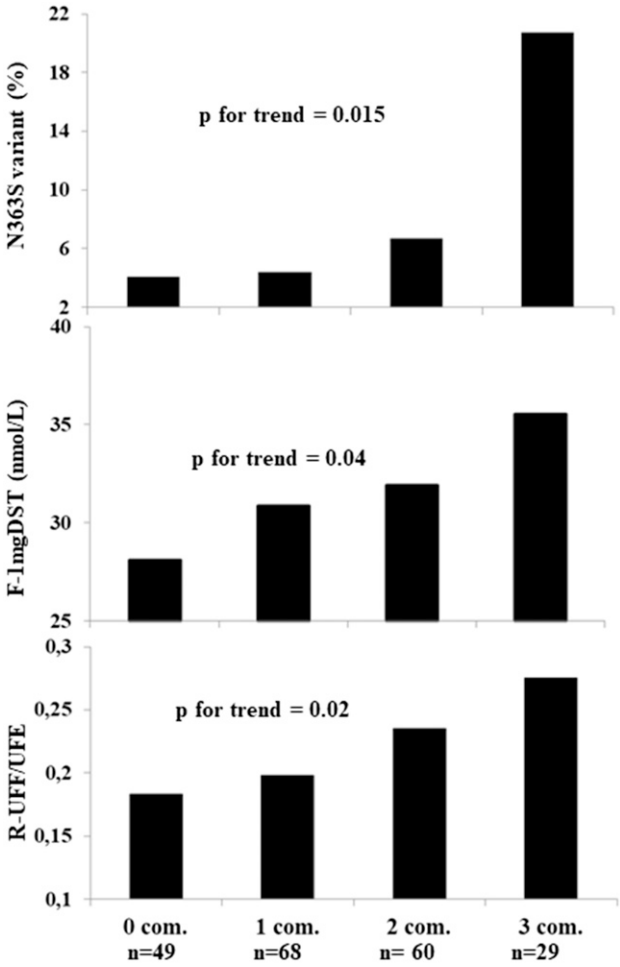

Figure 1. Association between GC secretion (as measured by F-1mgDST, bottom panel), GC peripheral activation (as measured by R-UFF/UFE, middle panel), and GC sensitivity (as mirrored by the *GR* gene N363S polymorphic variant, top panel) with the number of possible GC-related consequences among patients with hypertension, T2D, and fragility FX. Normeal F-1mgDST: <1.8 μg/dL (50 nmol/L). Normal UFF: 3 to 43 μg/24 hours (8.3 to 118.7 nmol/24 hours). Normal UFE: 15 to 122 μg/24 hours, (41.6 to 337.9 mol/24 hours). Abbreviation: com, possible consequences of cortisol excess among patients with hypertension, T2D, and fragility FX.

cortisol secretion is associated with hypertension, it is possible to hypothesize that the widely accepted cutoff of F-1mgDST set at 1.8 μg/dL (50 nmol/L) could be considered too high and, therefore, not sensitive enough for detecting patients with a possible cortisol-related cardiovascular risk, as was previously suggested in patients with subclinical hypercortisolism (33) and nonfunctional adrenal tumors (34). Second, in the future, the simultaneous assessment of GC secretion, GC peripheral activation, and GC sensitivity could be considered among the tools for detecting patients at risk for hypertension, T2D, and fragility FX.

The current study has some limitations. Its cross-sectional design allows the detection of highly significant associations but no causal relationship. However, a possible causative role for the slight defect of GCS, GC sensitivity, and GC peripheral activation appears plausible, given the well-known negative role of cortisol excess on bone, glucose metabolism, and blood pressure. Second, this study was designed to compare GCS, GC peripheral activation, and GC sensitivity in patients with T2D compared with matched patients without T2D. Therefore, given that patients with T2D *per se* displayed increased cortisol secretion, we could not completely exclude the possibility that the original study design might have biased the results, particularly considering the relatively small sample size. Indeed, the statistically significant difference in age- and BMI-adjusted F-1mgDST levels between patients with and those without hypertension was lost when patients with T2D were excluded from the analysis (Table 2). In our opinion, this lack of a statistically significant difference may be due to the reduced sample size. The fact that in the entire study group, F-1mgDST levels were statistically associated with hypertension even after adjustment for age, BMI, and T2D reinforces the idea that hypertension is associated with a slight defect in GCS. On the other hand, the finding that, at variance with F-1mgDST, the age- and BMI- adjusted difference in R-UFF/UFE levels between patients with hypertension and those without was confirmed even after exclusion of patients with T2D from the analysis might suggest that in eucortisolemic patients hypertension may be more strongly associated with GC peripheral activation (as mirrored by R-UFF/UFE) than with cortisol secretion (as reflected by GCS). This is in keeping with the well-known role of 11βHSD1 in amplifying GC action in cells and contributing to hypertension through direct and indirect effects on the kidney and vasculature (35). This hypothesis could explain our finding of a higher OR for predicting hypertension of R-UFF/UFE (125.9) compared with that of F-1mgDST (3.29). However, we emphasize that the independent associations between R-UFF/UFE levels and either hypertension or the simultaneous presence of T2D, hypertension, and fragility FX are somewhat weakened by the large CIs of R-UFF/UFE levels (3.29 to 4909.4 and 2.3 to 7417.8, respectively). In our opinion, these large CIs may be due to a huge statistical dispersion in a relatively small sample size.

In addition, we did not measure plasma dexamethasone concentration. This is a further limitation of the study because the results of the F-1mgDST are highly variable as a result of the between-individual variability in dexamethasone metabolism (1). Finally, we include only white patients to avoid possible biases related to the effect of the different races on vitamin D metabolism, bone, and vascular health (20). Therefore, the present

Table 3. Comparisons of Cortisol Secretion and Sensitivity and 11βHSD Type 2 Activity in Whole Patient Sample by Absence or Presence of Comorbidities Possibly Associated With Cortisol Levels in Patients With Hypertension, T2D, and Fragility Fx

Variable	No Comorbidities (n = 49)	One Comorbidity (n = 68)	Two Comorbidities (n = 60)	Three Comorbidities (n = 29)
Age, y	62.0 ± 8.5 (37–80)	65.0 ± 8.1 (50–80)	65.6 ± 7.4[a] (52–80)	68.6 ± 6.9[b] (54–78)
BMI, kg/m^2	27.5 ± 4.1 (19.9– 37.4)	29.2 ± 5.0 (20.9–39.8)	29.9 ± 5.0 (21.3–40.0)	29.3 ± 4.4 (21.0–40.0)
F-1mgDST, μg/dL[c]	0.99 ± 0.37 (0.4–1.8)	1.13 ± 0.44 (0.4–1.8)	1.16 ± 0.42[b] (0.4–1.8)	1.37 ± 0.45[b] (0.5–1.8)
R-UFF/UFE[c]	0.19 ± 0.08 (0.11–0.40)	0.22 ± 0.08 (0.07–0.50)	0.22 ± 0.10 (0.06–0.59)	0.26 ± 0.15[b] (0.11–0.51)
N363S heterozygous variant	2 (4.1)	3 (4.4)	4 (6.7)	6[b,d,e] (20.7)

Data are mean ± SD (range) or absolute number (%). Normal F-1mgDST: <1.8 μg/dL.

P for trend: F-1mgDST 0.001; *R*-UFF/UFE 0.009.

[a] $P < 0.02$ vs no comorbidities.

[b] $P < 0.05$ vs no comorbidities.

[c] After adjustment for age by general linear model.

[d] $P < 0.05$ vs one comorbidity.

[e] $P < 0.05$ vs two comorbidities.

findings concern only white persons. Overall, these considerations suggest that our findings should be taken with caution and should be confirmed in larger populations of patients with hypertension.

In conclusion, the current study suggests that in postmenopausal women without hypercortisolism, hypertension was independently associated with a slight defect in GCS and GC peripheral activation, and the number of possible consequences of cortisol excess among patients with T2D, hypertension, and fragility FX and their simultaneous presence are associated with GCS, GC peripheral activation, and GC sensitivity.

Further studies in a larger sample of patients are needed to confirm the present findings and to assess whether the risk of developing hypertension, T2D, and fragility fracture could be reduced by modulating cortisol secretion and GC peripheral activation.

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